Review Article

Current Pharma Research

ISSN: 2230-7842

CPR 3(3), 2013, 939-956.

Oro-Mucoadhesion-A Theoretical Overview.

***[1]A. A. Deshmukh, K. S. Salunkhe, S.R. Chaudhari.**

[1]Department of Pharmaceutics, Amrutvahini College of Pharmacy, University of Pune, Sangamner, Maharashtra, India.

Abstract

The oral cavity is fascinating site for delivery of many drugs since antique time. Nonetheless conventional dosage forms depicts lack of significant correlation between membrane permeability, absorption & bioavailability due to extensive presystemic clearance in liver followed by oral administration. Tribulations associated with conventional per-oral drug delivery & parenteral delivery became prerequisite for research of alternative routes for delivery of such drugs. These include various mucoadhesive systems predominantly. It had been the subject of great interest nowadays because mucoadhesion could be a solution for bioavailability problems by prolonging the residence time of the dosage form. Over the last 30 years, the market share of transmucosal drug delivery system has significantly increased with an estimated value of $6.7 million in 2006[3]. This overview enlighten briefly by discussing the detailed concept of mucoadhesion including mucoadhesive forces, various theories of mucoadhesion, mechanism of oro-mucoadhesion, anatomy of oral mucosa along with in-vitro/Ex-vivo & In vitro techniques.

Key Words

Oro-mucoadhesion, mucoadhesive forces, Transmucosal, buccal, ex-vivo.

Introduction

Traditionally, per-oral delivery has been the primary route of administration of therapeutic agents [l4]. Orally administered drugs shows major impediments as extensive first pass metabolism, poor drug bioavailability and stability problems in the gastrointestinal environment like instability in gastric pH & complexation with mucosal membrane. These hindrances can be overcome by altering the route of administration as parenteral, transdermal or transmucosal[15].

***Corresponding Author:**

deshmukh.anuradhaa@gmail.com

Intricacies associated with parenteral drug delivery have opened a new research platform for mucoadhesive drug delivery systems in recent years.

Over the last two decades, mucoadhesion has become an interesting topic for its potential to optimize localized drug delivery, by retaining dosage form at the site of action or systemic delivery, by retaining a formulation in intimate contact with the absorption site [11].

Muco / Bio Adhesion

Concept of Muco/ Bioadhesion

The concept of mucoadhesives was introduced into the controlled drug

delivery area in the early 1980's. American Society of Testing & Materials (1984) defined the term 'adhesion' as the state in which two surfaces are held together by interfacial forces which may consists of valence forces, interlocking action or both[7]. Mucoadhesives or bioadhesives can be defined as 'substance capable of interacting with biological material & being retained on them or holding them together for extended periods of time'[7]. Bioadhesion is the phenomenon between two materials, which are held together for extended periods of time by interfacial forces. It is generally referred as bioadhesion when interaction occurs between polymers & epithelial surface; mucoadhesion when occurs with the mucus layer covering a tissue. Generally Bioadhesion is deeper than mucoadhesion. However, these terms seems to be used interchangeably. [22]

Types of Mucoadhesive Systems[23]

Mucoadhesive systems can be classed based on potential site of attachment as: Buccal, Sublingual, Vaginal, Rectal, Nasal, Occular & gastrointestinal delivery systems. Among the various transmucosal routes rectal, vaginal & ocular delivery system shows poor patient acceptability. These systems are mainly restricted to delivery of drugs for local release the systemic drug delivery. Though Nasal drug delivery is now showing the clinical uses, small volume of the nasal cavity, rapid clearance of administered substances & potential disruption of physiological functions of nasal cavity proposes limitations of Nasal delivery. Also, nasal drug delivery is not feasible for chronic condition treatment as long term administration of drugs across nasal mucosa can cause irreversible damage to the nasal cilia. Whereas the advent of excellent accessibility, presence of smooth muscle & relatively immobile mucosa, buccal mucosa upsurge as a suitable candidate for administration of retentive dosage forms among the various transmucosal routes [Thornhill].

Need of Mucoadhesives[6]

Mucoadhesives plays vital role in prolongation of therapeutic effect by prolonging the contact time with the site. It can also be used for targeted & localized drug delivery. Mucoadhesives are used extensively for Controlled release formulations. It can improve the bioavailability by by-passing first pass metabolism, avoiding drug degradation & thus minimizing the dosing frequency & therapeutically effective dose of a drug & hence improvement in patient compliance. Mucoadhesive formulations can exhibits high drug flux through the absorbing tissue & reduction in fluctuations of steady state plasma level. Mucoadhesive drug delivery systems have opened a promising platform for newer researches like antihypertensives, anti-anginal, analgesics, anti-inflammatory, and anti-asthmatic, anti- infective, anti-neoplastic, hormonal & ophthalmic drugs.

Advantages of Mucoadhesives [2,20,17]

There are several advantages of mucoadhesive formulation which are as follows,

1. Improvement in Bioavailability due to direct entry of drug into systemic circulation by bypassing gastro-intestinal tract & hepatic portal system, protection of drugs from degradation due to pH & digestive enzymes of middle Gastro-intestinal tract (except gastrointestinal mucoadhesive formulations)
2. Low enzymatic activity than other oral routes (except for gastrointestinal mucoadhesive formulations)
3. Improved patient compliance due to elimination of pain in case of injections, administration of drugs to unconscious & incapacitated patients, convenience of administration as compared to injections or other conventional oral medications.
4. Controlled & Sustained drug delivery is possible & relatively rapid onset of action.
5. Use of permeation enhancers, enzyme inhibitors & pH modifiers in the formulation without observing permanent damaging effect on the mucosa.
6. Increased ease of drug administration & easy termination of therapy if therapy is to be discontinued (except for gastrointestinal)
7. As in Transdermal Drug Delivery Systems, lack of major barrier layer stratum corneum, so faster onset of therapy
8. Significant reduction in dose hence dose related side effects are minimized.
9. It offers comparatively shorter treatment period.
10. It offers a passive system of drug absorption & does not require any activation.
11. This route provides an alternative for administration of various hormones, narcotic analgesics, steroids, enzymes & cardiovascular agents etc.
12. Increased safety margin of high potency drugs due to better control of plasma levels
13. Reduction in fluctuations in steady state levels & therefore better control of disease condition & reduced intensity of local or systemic side effects.
14. Versatility in designing of dosage form as multidirectional & unidirectional release systems for local or systemic actions etc. creating new commercial & clinical opportunities for delivering narrow absorption window drugs at the target site to maximize their usefulness.

Limitations[2, 17]

In spite of various advantages mucoadhesive drug delivery systems shows some limitations also which are pointed as below

1. In case of gastro-intestinal mucoadhesive systems, limited gastric residence time which ranges from few minutes to 12 hrs. led to unpredictable bioavailability & time to achieve maximum plasma level
2. Drug administration via the buccal mucosa pose many problems such as low permeability, pH stability problems, various barriers for penetration of drug, enzymatic barriers, drugs which are irritant

to oral mucosa, bitter or unpleasant taste, odour cannot be administered by this route

3. Swallowing of formulation by the patient may be possible
4. Over hydration may lead to the formation of slippery surface & structural integrity of the formulation may get disrupted by the swelling & hydration of the bioadhesive polymer.

For development of efficient oro-mucoadhesive drug delivery system it is very important to understand the pharmaceutical considerations of the Oro-mucoadhesion.

Muco / Bioadhesive forces[5, 13, 17, 21,22]

Various forces take part in the phenomenon of Mucoadhesion which can be enlisted as: 1) Van der Waals forces; 2) Hydrogen bonding; 3) Disulphide Bridging; 4) Hydration forces; 5) Electrostatic double layer forces; 6) Hydrophobic interactions; 7) Steric forces; 8) Covalent bonds etc. Process of Bioadhesion may involve either Physical or Chemical interactions. Forces involved in Physical &/ or Chemical interactions are detailed in below table 1.

Factors affecting Muco / Bio adhesion[5, 8, 9, 10, 12, 14,17]

Many parameters such as Polymer related factors, Physiological factors & Environment related factors plays an important role in phenomenon of mucoadhesion. Detailed description of these factors is given in below table 2.

Mechanism of Bioadhesion[5, 21, 22]

For process of Bioadhesion various steps occur in progressions which are as follows:

Step I: Contact & Consolidation stage:

1] Spreading, wetting, swelling & dissolution of the mucoadhesive polymer at the mucus Interface.

2] Initiation of the intimate contact between the polymer & the mucus layer at the Interface

Step II: Interpenetration & Entanglement of bioadhesive material into the mucus layer

3] Inter-diffusion & interpenetration between the chains of the mucoadhesive polymers & the mucus gel (glycoprotein) network, creating a greater area of contact by physical cross links & mechanical interlocking. Strength of these bonds depends on degree of penetration between two polymer groups.

4] Orientation of the polymers at the interface by adsorption leading to entanglement & formation of secondary chemical bonds between polymer chains & the mucin molecules.

Theories of Muco / Bioadhesion[17, 19]

The complex phenomenon of Mucoadhesion involves various physical & chemical interaction bonds which were elaborated earlier in section 1.1.3. Based on these, till date six theories were proposed to explain the phenomenon of Mucoadhesion which is exemplified in below table 3.

Oro-Mucoadhesion

Anatomy of Buccal Mucosa & its suitability[1, 6, 12, 16, 18, 22]

As discussed earlier in point 1.2, oral cavity is the novel & proficient site for drug delivery. Drug delivery via the membranes of the oral cavity can be classed as:

- Sublingual delivery: involves administration through the membranes of the ventral surface of the tongue & the floor of the mouth to the systemic circulation. Generally employed for the delivery of drugs characterized by a high permeability across the mucosa & used in the treatment of acute disorders.
- Buccal delivery: involves administration through the buccal mucosa, mainly composed of the lining of the cheeks. Generally used in treatment of chronic disorders when a prolonged action of active substance is required.
- Local delivery: consisting of administration through all areas other than former two regions that is palate, gingival or cheek.

Anatomy of buccal mucosa

The epithelium is similar to stratified squamous epithelia found in rest of the body & is about 40-50 cell layers thick. Lining epithelium of the buccal mucosa is the non-keratinized stratified squamous epithelium that has thickness of approx. 500-600 µ & surface area of 50.2 cm^2. Basement membrane, lamina propria followed by the submucosa is present below the epithelial layer. Lamina propria is rich with blood vessels & capillaries that open to the internal jugular vein.

The primary function of buccal epithelium is the protection of the underlying tissue. In non-keratinized regions, lipid-based permeability barriers in the outer epithelial layers protect the underlying tissues against fluid loss & entry of potentially harmful environmental agents such as antigens, carcinogens, microbial toxins & enzymes from foods & beverages.

Barriers to penetration across buccal mucosa[5, 17, 22]

Saliva, mucus, membrane coating granules, basement membrane etc. act as major barriers for penetration across buccal mucosa which retards the rate & extent of drug absorption through the buccal mucosa. The main penetration barrier exists in the outermost quarter to one third of the epithelium.

In general, intercellular spaces serves as major barrier for permeation of lipophilic compounds & lipophilic cell membranes for hydrophilic compounds due to low partition coefficients.

Membrane coating granules (MCG's)

These are also known as keratinosomes, cementsomes, transitory dense bodies, 'small spherically shaped granules' etc.

Description: Spherical or oval shaped having diameter of 100-300 nm & found in both keratinized & non-keratinized epithelia. Permeability barrier property of the mucosa is predominantly due to intercellular materials derived from MCGs.

Occurrence: Found near upper, distal or superficial borders of cells & few occur near opposite border.

Function: Membrane thickening effect, Cell adhesion, production of cell surface coat, cell desquamation & permeability barrier.

Mechanism for function of barrier: MCGs discharge their contents into the intercellular space to ensure epithelial cohesion in the superficial layers, & this discharge forms a barrier to the permeability of various compounds.

Basement membrane

Basement membrane also plays a role in limiting the passage of materials across the junction between epithelium & connective tissue.

Mechanism: Similar mechanism as that of MCGs appears to operate in the opposite direction. Charge on the constituents of basal lamina may limit rate of penetration of lipophilic compounds that can pass through superficial epithelial barrier relatively easily.

Saliva: It is an unstirred layer providing approx. 70 µm thick salivary coating on mucosal surface. Saliva contains high molecular wt. mucin named MG1 that can bind to surface of oral mucosa so as to maintain hydration, provide lubrication, concentrate protective molecules such as secretory immunoglobulins & limit the attachment of micro-organisms. Saliva contains enzymes in moderate levels e.g. esterases, carbohydrases, phosphatases, aminopeptidases & various proteolytic enzymes etc. which acts as enzymatic barrier for penetration through buccal mucosa. The use of mucoadhesive polymers as enzyme inhibitor agents has been developed to overcome this obstacle in peptide & protein delivery.

Mucus: The epithelial cells of buccal mucosa are surrounded by the intercellular ground substance called mucus with thickness varying from 40 µm to 300 µm. Sublingual glands & minor salivary gland together produce the majority of mucus & is secreted by goblet cells lining the epithelia or by special exocrine glands with mucus cells 'acini'.

Composition of mucus: Mucus is composed chiefly of mucins & inorganic salts suspended in water. The exact composition of mucus layer varies substantially, depending on the species, anatomical location & normal or pathological state of organism. Composition of mucus is depicted in below table 4.

Primary functions of mucus layer are:

Protective	Results particularly from its hydrophobicity that protects the mucosa from luminal diffusion of hydrochloric acid to epithelial surface.
Barrier	It possesses a diffusion barrier for molecules & especially against reabsorption. Physicochemical properties such as molecular weight, molecular charge, hydration radius & ability to form hydrogen bonds etc. influences diffusion of molecules through the mucus layer.
Adhesion	It has strong cohesive properties & firmly binds to epithelial cells surface as continuous gel layer.
Lubrication	An important role to keep the layer moist due to their viscous gel forming properties & general stickiness.

Muco-adhesion	At physiological pH, the mucus network may carry a significant negative charge because of the presence of sialic acid & sulphate residues & this high charge density due to negative charge contributes significantly to bioadhesion.

At buccal pH, mucus can form a strongly cohesive gel structure that binds to epithelial cell surface as a gelatinous layer. Mucus molecules are able to join together to make polymers or an extended three dimensional network.

Absorption Pathways[22]

Determination of Mucoadhesion[5,7]

Mucoadhesion can be determined using various In-vitro/Ex-vivo & In vivo techniques. These techniques are summarized in below table 5.

Conclusion

In conclusion, bioadhesion is a great area of interest to improve & enhance the bioavailability of drug by prolonging residence time of dosage form onto absorption surface. It can be adapted to almost all of the administration routes for local as well as systemic effects. Process of mucoadhesion is a very complex phenomenon involving wetting, swelling of bioadhesive material onto mucous layer followed by Interpenetration and entanglement of material into mucus layer and formation of chemical bonds. Various forces & bonds interact in process of bioadhesion along with various factors such as polymer related, environment related & physiological factors. Oro-Mucoadhesive drug delivery system shows a very good potential as alternative to overcome the limitations of conventional drug delivery & parenteral administration. Improvements in bioadhesive based drug delivery particularly the delivery of novel; highly effective & mucosa friendly polymers are creating new commercial & clinical opportunities for delivering narrow absorption window drugs at the target site to maximize their usefulness. Various mucoadhesive polymers, enzyme inhibitors, & penetration enhancers are used in the formulation to overcome the barriers posed by oral mucosa. It will continue be an exciting research platform for improving drug bioavailability & thus increased patient compliance. Although palatability, irritancy & formulation retention at site of application need to be considered in the design of such formulation.

References

1. Alagusundaram M, Chetty C M, Dachinamoorthi D. Advances in buccoadhesive drug delivery system – A review. Int. J. Rev. in Life Sci. 2011; 1(2): 35-44.

2. Alexander A, Ajazuddin, Tripathi DK, Verma T, et al. Mechanism responsible for Mucoadhesion of Mucoadhesive Drug Delivery System: A Review. Int. J. App. Bio. & Pharm. Tech. 2011; 2(1): 434-445.

3. Andrews G.P., Laverty T. P., Jones D. S. Mucoadhesive polymeric platforms for

controlled drug delivery. Eur. J. Pharm. and Biopharm. 2009; (71): 505–518.

4. Bhalodia R, Basu B, Garala K, Joshi B, et al. Buccoadhesive Drug Delivery Systems: A Review. Int. J. Pharma & Biosciences 2010; 1(2): 1-32.

5. Das R, Ali K A, Bandyopadhyay A K. Mucoadhesion & Mucoadhesive Tablets- A Review. Int. J. Pharm. Sci. Tech 2011; 6(1): 64-115.

6. Gupta S K, Singhvi I J, Shirgat M, Karwani G, et al. Buccal adhesive drug delivery system: A review. Asian J. Bio. & Pharm. Res. 2011; 2(1): 105-114.

7. Khar R. K., Ahuja A. and Ali J., Mucoadhesive Drug Delivery; 1st ed.; N. K. Jain Ed.; Controlled and Novel Drug Delivery; C.B.S. Publishers and Distributors: New Delhi; 2007; 16: 353-380.

8. Kast C E, Bernkop-Schnürch A. Thiolated Polymers-thiomers: Development & in vitro evaluation of chitosan-thioglycolic acid conjugates. Biomaterials 2001; 22: 2345-2352.

9. Langoth N, Kalbe J, Bernkop-Schnürch A. Development of Buccal Drug Delivery Systems based on a thiolated polymer. Int. J. Pharm. 2003; 252: 141-148.

10. Marschütz M K, Bernkop-Schnürch. Thiolated Polymers: Self-crosslinking properties of thiolated 450 kDa poly(acrylic acid) & their influence on mucoadhesion. Eur. J. Pharm. Sci. 2002; 15: 387-394.

11. Murali krishna K,Nagaraju T, Gowthami R, Rajashekar M, et al. COMPREHENSIVE REVIEW ON BUCCAL DELIVERY. Int. J. Pharm. 2012; 2(1): 205-217.

12. Patel M, Karigar A, Savaliya P, Ramana MV, et al. Buccal Drug Delivery: The Current Interest. Int. Res. J. Pharm. 2011; 2(12): 4-11.

13. Peppas N A, Thomas J B, Mcginity J. Molecular aspects of Mucoadhesive Carrier Development for Drug Delivery & Improved absorption. NIH-PA Author Manuscript PMC 2011: 1-26.

14. Punitha S, Girish Y. Polymers in mucoadhesive buccal drug delivery system – A review". Int. J. Res. Pharm. Sci. 2010; 1(2): 170-186.

15. Puratchikody A, Prasanth V.V, Mathew S T, Ashok Kumar B. Buccal Drug Delivery: Past, Present and Future – A Review. Int. J. Drug Deli. 2011; 3: 171-184.

16. Pather S I, Rathbone M J and Senel S, Oral Transmucosal Drug Delivery; 2nd ed., Rathbone M.J., Hadgraft J, Roberts M.S., Lane M. E Ed.; Modified–Release Drug Delivery Technology; Informa Healthcare: New York, 2008; 1, 5: 53-73.

17. Reddy P C, Vishnu Y V, Rao Y

M, Buccal Drug Delivery System; Rao Y M, Jithan A V, Ed.; Advances in Drug Delivery; PharmaMed Press: Hyderabad, 2011, 1, 3: 139-212.

18. Rossi S, Sandri G, Caramella C M. Buccal Drug Delivery: A Challenge already won?. Drug Disc. Today Tech. 2005; 2(1): 59-65.

19. Roy S K, Prabhakar B. Bioadhesive polymeric platforms for Transmucosal Drug Delivery Systems – a Review. Trop. J. of Pharma. Res. 2010; 9(1): 91-104.

20. Sharma S, Alexander A, Swarna, Ajazuddin. Theories & factors affecting Mucoadhesive Drug Delivery Systems: A Review. Int. J. Res. Ayur. & Pharm. 2011; 2 (4): 1155-1161.

21. Smart J D. The Basics and underlying mechanisms of mucoadhesion. Adv. Drug Deli. Rev. 2005; 57: 1556-1568.

22. Sudhakar Y, Kuotsu K, Bandyopadhyay A.K. J. Contr. Rel. 2006; 114: 15–40.

23. Thornhill M H, Hearnden V, Sankar V, Hull K. New Adv. Drug Deli. Rev. 2012, 64: 10-28.

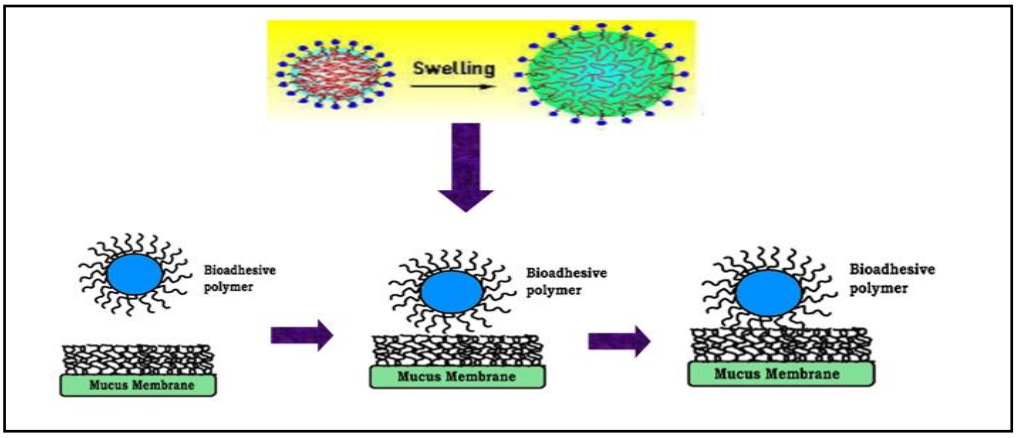

Fig. 1: Contact Stage & Consolidation Stage-Spreading, Wetting & Swelling.

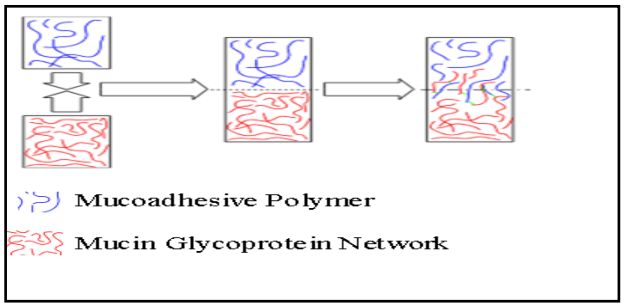

Fig. 2: Inter-diffusion & interpenetration interactions.
The Inter-penetration Theory; three stages in the interaction between a mucoadhesive polymer & mucin glycoprotein.

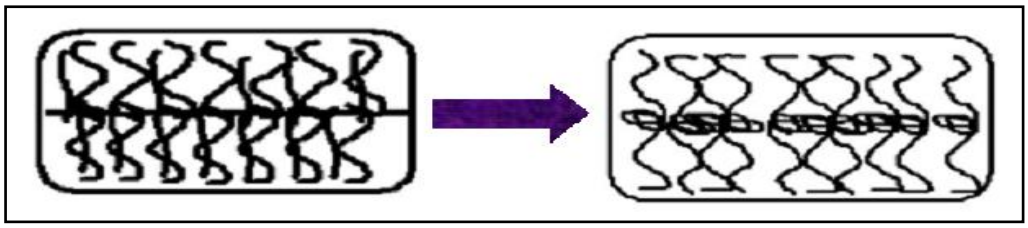

Fig. 3: Formation of chemical bonds.

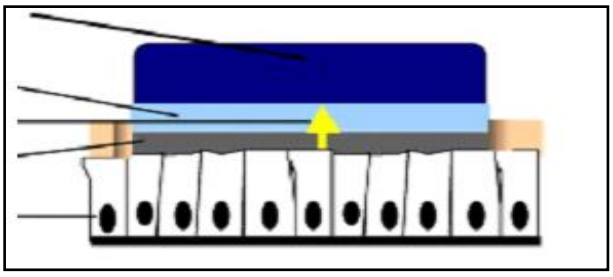

Fig. 4: Wetting Theory.

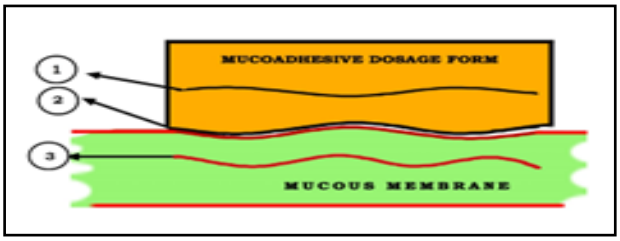

Fig. 5: Fracture Theory.

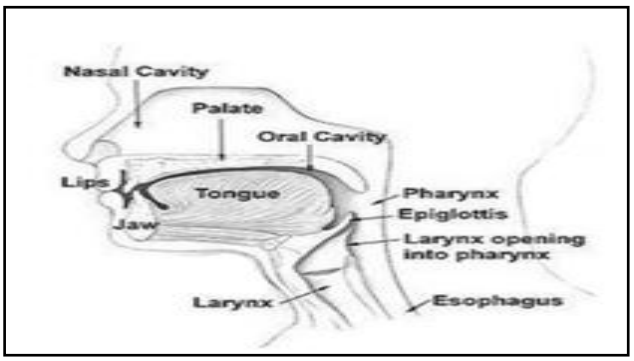

Fig. 6: Oral Cavity.

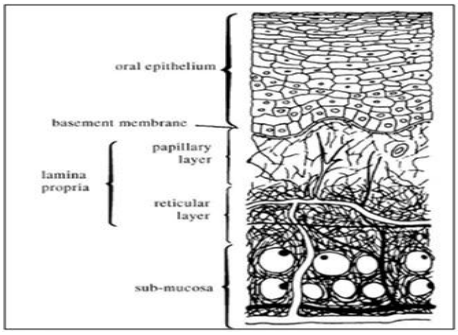

Fig. 7: Buccal Mucosa.

Table 1: Forces Responsible For Bio / Mucoadhesion.

Physical interaction forces:	**Van der waals force**	London Dispersion Forces (Dispersion forces): These occur due to electronic motions in paired molecules & involve the interaction between temporarily induced dipoles in non-polar molecules. These interactions involve a force of about 0.5 – 1 K cal/mole.
		Dipole-dipole interactions (Keesom interactions): These occur with two molecules having permanent dipoles. This interaction involves a force of 1-7 K cal/mole.
		Debye type forces: These are involved in interactions between permanent & induced dipoles. These interactions involves a force of about 1-3 K cal/mole.
	Hydrogen bonds	Weak bonds formed between an H atom (having a slightly positive charge) covalently attached to an electronegative atom & another electronegative atom, although transfer of electrons do not occur. E.g.: Formation of gelled structure during the mixing of aqueous solutions of polyvinyl alcohol & glycin.
	Hydrophobic bonds	Formation of bonds due to interaction of non-polar groups of polymers dispersed in an aqueous solution. Water molecules adjacent to non-polar groups form hydrogen bonded structures thereby lowering the system entropy & hence increasing the tendency of non-polar groups to associate with each other to minimize this effect. E.g.: Freeze thawing of polyvinyl alcohol solution in water.
Chemical interaction forces:	**Ionic bonds**	These are the strong bonds formed amongst the polymers when two oppositely charged ions attract each other via electrostatic interactions. E.g.: Instantaneous formation of gelled structure when alginate & chitosan are mixed in water.
	Covalent bonds	These are strong bonds formed due to sharing of electrons in pairs amongst the bonded atoms. E.g.: Cross linking reaction between genipin & amino groups

Table 2: Factors Affecting Bio / Mucoadhesion.

Factors affecting Muco / Bioadhesion		**Comments**
Polymer related factors	Molecular weight of the polymer	In general threshold required for successful bioadhesion is at least 100,000 molecular weight. For a linear polymer bioadhesiveness improves with increasing molecular wt. Low molecular weight polymers can interpenetrate more easily, whereas entanglements are important for high molecular weight polymers.

	Concentration of the polymer used	This is an optimum concentration of a bioadhesive polymer to produce maximize bioadhesion. In case of high concentrated system, beyond the optimum level, however the adhesive strength drops significantly because the coiled molecules become separated from the medium so that the chain available for interpenetration becomes limited.
	Polymer Chain Length	Mucoadhesive property of a polymer increases with increase in chain length of a polymer.
	Spatial Conformation	Three dimensional structure of a polymer is important. In general, polymer with a helical conformation is able to shield adhesively active groups & therefore a much higher molecular mass is needed for the same adhesive strength as a linear polymer.
	Flexibility of Polymer chains	It is important for interpenetration & entanglement between polymer & mucosal layer. Cross linking of water soluble polymer decreases the mobility of individual polymer chain & hence decreases the effective chain length that can penetrate into the mucus layer, which in turn reduces bioadhesive strength.
	Hydrogen bonding capacity	For mucoadhesion to occur, desired polymers must have functional groups that are able to form hydrogen bonds.
	Cross linking density	The average pore size, the molecular weight of the cross linked polymer & the density of cross linking are the three important interrelated structural parameters of a polymer network. Increased cross linking density results in insufficient swelling of the polymer & decreased rate of interpenetration between polymer & mucin.
	Charge on polymer chain	Non-ionic polymers have smaller degree of adhesion compared to anionic polymers. Strong anionic charge on the polymer is one of the required characteristics of mucoadhesion.
	Swelling (Hydration)	Swelling is required for a mucoadhesive polymer to expand & create a proper "macromolecular mesh" of sufficient size & also to induce mobility in the polymer chains in order to enhance the interpenetration process between polymer & mucin. Over hydration results in the formation of a wet slippery mucilage without adhesion. Swelling characteristics are related to the bioadhesive itself & its environment. Swelling depends on the polymer concentration, ionic strength as well as the presence of water.
Physiological factors	mucin turn over	Mucin turnover produces substantial amounts of soluble mucin molecules that interact with mucoadhesives before they interact with mucus.
	Disease state	The physiology of the mucosal layer may vary based on the pathophysiological nature of the human body such as common cold, gastric ulcers, ulcerative colitis, cystic fibrosis, bacterial & fungal infections of the female reproductive tract, inflammatory conditions of the eye, tissue fibrosis allergic rhinitis etc.

	Tissue environment	Eating, drinking, talking, peristaltic movements & other GI movements in the GIT.
Environment related factors	pH	Change in pH of the microenvironment can alter the ionization state, and, therefore, the adhesion properties of a polymer, as differences in dissociation of functional groups on the carbohydrate moiety & the amino acids of the polypeptide backbone of the mucus.
	Applied strength	Initial pressure applied to the site of contact affects the depth of interpenetration of polymer chain. The adhesion strength increases with the applied strength or with the duration of its application, up to an optimum. If high pressure is applied for a sufficiently long period of time, polymers become mucoadhesive even though they do not have attractive interaction with the mucin.
	Initial contact time	Bioadhesive strength increases as the initial contact time increases. It determines the extent of swelling & the interpenetration of polymer chains.
	Selection of the model substrate surface	Bioadhesive substrates plays an important role. Physical & biological changes may occur in mucus gels or tissues under experimental conditions.
	Presence of metal ions	Metal ions interact with charged group of polymers &/or mucus thereby decreasing the number of interaction sites & the tightness of mucoadhesive bonding.

Table 3: Theories & Mechanisms of Mucoadhesion.

Theory	**Mechanism of bioadhesion**	**Comments**
Electronic theory:	Attractive electrostatic forces between glycoprotein mucin network and the bioadhesive material.	Electrons transfer occurs between the two forming a double layer of electric charge at the surface. Adhesion occurs due to attractive forces across the double layer.
Wetting theory: **1.** Mucoadhesive dosage form **2.** Hydrating region in dosage form **3.** Direction of water movement **4.** Dehydrated Mucus Layer **5.** The Mucosa	Ability of bioadhesive polymer to spread and develop intimate contact with the mucous membrane.	Predominantly applicable to liquid bioadhesive systems. Spreading coefficient of polymers must be positive. Contact angle between polymer and cells must be near to zero.
Adsorption Theory:	Surface forces resulting in chemical bonding.	**Strong primary force**: covalent bonds which are undesirable as their high strength may result in permanent bonds. **Weak secondary forces**:

		hydrogen bonds, hydrophobic bonds, Electrostatic forces & Van der Waals forces. In adhesion, primary bonds results because of chemisorptions due to ionic, covalent & metallic bonding & secondary bonds arise mainly because of van der waals forces, hydrophobic interactions & hydrogen bonding.
Diffusion theory: Refer Fig. 2	Physical entanglement of mucin strands and flexible polymer chains.	For maximum diffusion and best adhesive strength, solubility parameters of the bioadhesive polymer and the mucus glycoproteins must be similar. Depth of penetration depends on diffusion coefficient & time of contact. Diffusion coefficient depends on molecular weight between cross links & decreases significantly as increase in cross linking density.
Mechanical Theory:	Adhesion arises from an interlocking of liquid adhesive into irregularities on the rough surface.	Rough surfaces provide an increased surface area available for interaction along with an enhanced viscoelastic and plastic dissipation of energy during joint failure, which are more important in the adhesion process than a mechanical effect.
Fracture theory: 1. Fracture at the mucoadhes Dosage form; 2. Fracture at the Dosage For Mucous Interface **3.** Fracture at the Mucous Membran	Analyses the maximum tensile stress developed during attachment of the transmucosal DDS from the mucosal surface. Relates the adhesive strength to the forces required for detachment of two involved surfaces after adhesion.	Does not require physical entanglement of bioadhesive polymer chains and mucous strands, hence it is appropriate to study bioadhesion of hard polymers which lack flexible chains.

Table 4: Composition of Mucus.

Constituents	**Amount (% w/w)**
Water	95
Glycoprotein & lipids	0.5-5.0
Mineral salts	1
Free proteins	0.5-1.0

Table 5: Absorption Pathways.

Absorption Pathways	**Comments**	
1. Passive diffusion: ▪ Transcellular route:	Also called as Intercellular route. The route involves material crossing the cell membrane & entering the cell. - The flux of drug through the membrane under sink condition can be given as: $J_c = \frac{(1-\varepsilon)D_c K_c}{h_c} C_d$	Where, K_c= partition coefficient Between lipophilic cell membrane & aq. Phase D_c = Diffusion coefficient of drug in transcellular spaces h_c = path length of transcellular route C_d= Donar drug concentration
▪ Paracellular route:	Also called as intercellular route. It involves passage of materials between the cells - The flux of drug through the membrane under sink condition can be given as: $J_p = \frac{D_p \varepsilon}{hp} C_d$	Where, Dp = Diffusion coefficient of the permeate in intercellular spaces hp = path length of paracellular route ε = area fraction of the paracellular route C_d= Donar drug concentration
2. Carrier mediated transport	Lipid solubility & molecular weight of the diffusant influences absorption potential of the buccal mucosa. Some drugs shows increased absorption when carrier pH is lowered & decreased absorption with increase in pH.	
3. Endocytosis	In very few cases, the drug molecules were engulfed by the cells so as to lead absorption. Active transport processes does not operate within the oral mucosa; it is believed that acidic stimulation of the salivary glands with the accompanied vasodilation, facilitates absorption & uptake into circulatory systems.	

Table 5: Mucoadhesion Determination Techniques.

Technique	**Mechanisms**	**Comments**
✓ **In Vitro / Ex-vivo:**		
▪ Shear strength	It measures the force required to separate two parallel glass slides	Although reproducible results, the technique involves

	covered with the polymer & with a mucus film.	no biological tissue & therefore does not provide a realistic simulation of biological conditions.
▪ Tensile strength:		
- Wilhelmy Plate technique	It measures dynamic contact angles, hence measures the bioadhesive force between mucosal tissue & dosage form.	By using CANH software system, parameters such as fracture strength, deformation to failure & work of adhesion can be analyzed.
- Electromagnetic force transducer	It measures tissue adhesive forces by monitoring the magnetic force required to exactly oppose the bioadhesive force.	Unique identity to record remotely & simultaneously the tensile force formation as well as high magnification video images of bioadhesive interactions at near physiological conditions.
- Texture analyzer	It measures the force required to remove the formulation from a model membrane, which can be a disc composed of mucin.	The force required to detach the mucin disc from the surface of the tablet, the tensile work, the peak force & the deformation can be obtained from the force-distance curve.
▪ Fluorescent Probe method	The lipid bilayer & proteins of membrane are labeled with pyrene & fluorescein isothiocynate respectively. The cells were then mixed with candidate bioadhesive, & the changes in fluorescence spectra are monitored.	This gives a direct indication of polymer binding & its influence on polymer adhesion.
▪ Flow channel method	Humid air at 37 C is passed through a glass channel filled with 2 % (w/w) aqueous solution of bovine submaxillary mucin. Bioadhesive polymer is placed on the mucin gel.	The static & dynamic behavior can be monitored at frequent intervals using a camera.
▪ Mechanical spectroscopic method	Carri-Med CSL100 rheometer with a 4 cm parallel plate 0.5 mm gap for this study is used.	Spectroscopic methods can provide chain interpretation or formation of hydrogen bonds.
▪ Falling liquid film method	The adhesion of particles to a small intestinal segments from rats placed at an inclination of a tygon tube flute is monitored by passing the particles suspensions over the surface.	By comparing the fraction of particles adherent to the tissue, the adhesion strength of different polymers can be determined.
▪ Colloidal gold staining method	This technique employs red colloidal gold particles, which are stabilized by the adsorbed mucin molecule by forming mucin-gold conjugates.	Upon interaction with mucin gold conjugates, bioadhesive hydrogels develop a red colour on the surface that can be quantified, either by the measurement of the intensity

		of the red colour on the hydrogel surface or by the measurement of the decrease in the concentration of the conjugates from the absorbance changes at 525 nm.
▪ Viscometric method	The viscosity of a combination of mucin & polymer dispersion is measured by Brookfield viscometer.	Quantify mucin-polymer bioadhesive bond strength.
▪ Optical biosensor or resonant mirror biosensor technique	The molecules in solution, when binding to the immobilized molecules, alter the refraction index of the medium & this change is detected by the screening of a laser beam.	Measures the interaction between glycoprotein of the mucus & different polymers.
▪ Biacore test	The sensor is the chip with the glass surface covered in a fine gold layer, where functional groups are introduced & the polymer is attached.	This test is based on the passage of a mucin suspension through a sensor containing the immobilized polymer. When a mucin particle binds to the polymer at the sensor, both the solute concentration & the refractive index on the surface undergoes changes.
▪ Atomic force microscopy	Changes in surface topography are indicative of the presence of polymer bound onto buccal cell surfaces.	It can be used under any environmental conditions; in air, liquids or vacuum. It enlarges more than 109 fold, which enables visualization of isolated atoms & offers a three-dimensional image of the surface.
▪ Florescence microscopy & Confocal laser scanning microscopy	It is a technique for obtaining high resolution optical images with depth selectivity.	It offers better visualization of mechanisms involved in the mucoadhesion.
▪ Electromagnetic force transduction	In addition to information about bioadhesive forces, this technology also offers the simultaneous video image of the interactions, with high resolution and under physiological conditions.	--------

▪ A lectin binding inhibition technique	Involves an avidin-biotin complex & a colorimetric detection system was developed to investigate the binding of bioadhesive polymers to buccal epithelial cells without having to alter their physicochemical properties by the addition of their "marker" entities.	The lectin from Canavalia ensiformis (Concanavalin A) has been shown to bind sugar groups present on the surface of buccal cells. Therefore, if polymer binds to buccal cells, they would mask the surface glycoconjugates thus reducing or inhibiting Canavalia ensiformis lectin binding.
✓ **In vivo:**		
▪ Using radio isotopes	This involves use of radio-opaque markers e.g. - barium sulphate, encapsulated in bioadhesive DDS to determine the effects of bioadhesive polymers on GI transit time.	Mucoadhesive labeled with Cr-51, Tc-99m, In-113m, or I-123 has been used to study the transit of the DDS in the GI tract.
▪ Gamma scintigraphy	This gives information in terms of oral dosage forms across different regions of GI tract, time & site of disintegration of dosage forms, site of drug absorption & also the effect of food, disease & size of dosage form on the in-vivo performance of the dosage forms.	Various factors to be considered for studying behavior of solid dosage forms include selection of radio isotopes, radio labeling & choice of imaging device.
▪ Pharmaco-scintigraphy	It is a tool to examine drug delivery to the eye.	New technique need to be exploited to maximum for its potential in evaluation of new molecular entities, drug delivery systems and therapeutic drug monitoring.
▪ X-ray Studies	Testing in vivo adhesion of barium sulphate matrix tablet contains drug & polymer by X-ray.	Determines the mucoadhesion time in vivo.

******